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INFLUENCE OF MINE ACID ON PLANKTONIC ROTIFERS[1]

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Abstract. Planktonic rotifers were sampled in spring and summer from 2 lakes in southeastern Ohio. Unpolluted waters of Dow Lake were lower in sulfate and iron and higher in pH, hardness and alkalinity than acid mine polluted waters of Lake Hope. Rotifers of Dow Lake exceeded those of Lake Hope in species number, numerical abundance, species diversity, and faunal similarity among sample dates; similarity between lakes was negligible. *Keratella cochlearis* and *Brachionus urceolaris*, the most abundant rotifers in each of the respective lakes, both showed broad tolerances to pH. While *B. urceolaris* showed a large selective advantage at low pH, this was reduced and both species fared better at near-neutral pH. Neither species appeared sensitive to alternative pH conditions in preference tests.

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Mine effluent is an important source of pollution in coal producing areas of the eastern United States. Major environmental effects are:

1) reduction of pH in receiving waters by an increase in sulfate and hydrogen ions

2) increase in coal-associated metalic ions such as iron and manganese

3) smothering of stream beds with a yellowish ferric hydroxide precipitate.

Studies of fauna in lakes and streams polluted by mine drainage generally indicate fewer pollution-tolerant than pollution-sensitive species (Lackey 1938, Dinsmore 1968, Parsons 1968, Roback and Richardson 1969, Warner 1971) and reduced species diversity in polluted relative to unpolluted waters (Dills and Rogers 1974, Orciari and Hummon 1975).

Rotifers have received little attention with respect to acid mine pollution. They have been reported in streams polluted with mine acids, but until recently (Evans 1978) without quantitative data at the species level (Lackey 1938, Parsons 1968). Still less is known about the effects of mine acidity on lake-dwelling rotifers.

Correlation between ploimate rotifer species and hydrogen ion concentration, derived from organic sources, was recognized by Harring and Myers in 1928. Alkaline (hard) water tends to support a rotifer subcommunity with few species and many individuals, while acid (soft) water supports a greater number of species each with fewer individuals. Myers (1931) divided these rotifers into 3 groups: an alkaline fauna restricted to pH values above 7, an acid fauna restricted to pH values below 7, and a transcursion fauna tolerant of both acid alkaline conditions.

We decided that a survey of 2 matched lakes, one unpolluted and one polluted by mine effluents, would help delineate the influence of nonorganic acids on planktonic rotifers. Abundance, species diversity and faunal similarity were compared during 2 seasons. Levels of acute toxicity and behavioral preference to mine acids were determined for the 2 most important species in the study.

METHODS AND MATERIALS

Studies were made in 2 dendritic, earth-dam impoundments in southeastern Ohio. Dow Lake (Strouds Run State Park, Athens County), established in 1960, has 60 ha surface area, 11 km of wooded shoreline and no source of acid mine pollution. Lake Hope (Zaleski State Park, Vinton County), established in 1939, has 50 ha surface area, 8 km of wooded shoreline and receives effluent from nearly 150 mine sites. Three sampling sites were selected in the upper, middle and lower thirds of each lake, adjacent

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to the main flow-through current. Rotifers were sampled 3 times in late spring and twice in late summer of 1971.

At each sampling site, temperature and dissolved oxygen were determined for surface and 1 m depth intervals using a YSI Model 51 T–O Meter. Water samples, 2.5 ℓ (Dow Lake) or 10 ℓ (Lake Hope) were taken at the same depths with a Van Dorn bottle. Plankton were captured on a 64 μm sieve, washed into a vial, fixed with 10% formalin, and the volume brought up to 20 ml. A sample of sieved water was also retained for chemical analysis. On each sampling date, 3 depths were sampled at the upper, 5–6 at the middle and 7–8 at the lower site.

Rotifers were tallied as to species for each sample by whole Sedgwick-Rafter cell counts. The 5 replicates recommended by Kutkuhn (1958) represented 25% of the total. Chemical analyses were made within 6 hr of collection, pH being determined with an Instrumentation Lab Model 265 Electrometer. Calcium and total hardness, total alkalinity, sulfate and iron tests were made with Hach Chemical Co. water analysis kits. All data are presented as mean ± standard deviation.

Tolerance and preference tests were performed in the laboratory on the dominant rotifers *Keratella cochlearis* (Gosse 1851) from Dow Lake and *Brachionus urceolaris* (Muller 1773) from Lake Hope. Tolerance to hydrogen-ion concentration was tested by observing mortality through time in a series of solutions at integer intervals of pH 2 to 11, using 5 to 20 specimens of each species per test. Test solutions, prepared from filtered lake waters and sulfuric acid or calcium hydroxide, were allowed to stabilize over time without buffering. To prevent contamination, animals were transferred through a series of rinse dishes containing the appropriate solution before tests began. After 24 hr at 22 °C, animals not responding to tactile stimulation were considered dead. Native lake waters served as controls for the respective species. Experimental mortality was corrected to compensate for mortality among control animals.

Preference for hydrogen-ion concentration was tested by observing locomotor response over time to gradient pH conditions, using 10 of each species per test gradient, one at a time. In the apparatus used (Ganning and Wulff 1966), two similar or contrasting test solutions entered a small, elongate chamber from opposite ends at constant and identical pressures. Solutions mixed in the center of the chamber and exited together through a plankton-gauze lined slit in the bottom. Sharpness of gradient was confirmed periodically by injection of dilute food coloring. The chamber was small enough to be completely visible at 30× under a dissecting microscope and was evenly illuminated from the bottom. Test solutions were of pH 3, 5, 7 and 9, and Dow Lake vs Lake Hope water, all gradient and non-gradient permutations being tested. Animals placed in the gradient zone were observed for 10 min, and number of gradient encounters, choices made after each encounter, and time between encounters were recorded.

RESULTS

Lake temperatures increased from mid-teens to mid-twenties in late spring and dropped from upper-to lower-twenties in late summer. Thermoclines developed in late spring, but temperatures were rather homogeneous in late summer. Chemical compositions of the two lakes during sampling periods are compared in table 1. Dissolved oxygen, sulfate and iron were less in Dow Lake than in Lake Hope samples, as analysed by means tests, whereas pH, hardness and total alkalinity were greater.

TABLE 1

Comparison of Chemical Composition between Dow Lake and Lake Hope.

Chem. Compos. (ppm)	Dow Lake	Lake Hope	Level of Signif.
Dissolved O_2	8.4± 1.9	9.1± 1.4	.01
Sulfate	52.0± 5.0	68.0±41.0	.001
Iron	0.1± 0.2	0.4± 0.2	.001
Calcium hardness	79.0± 8.0	48.0±10.0	.001
Total hardness	118.0±21.0	87.0±18.0	.001
Total alkalinity	77.0±12.0	17.0± —	.001
pH	8.0± 0.5	4.0± 0.2	.001

*All values except significance mean±standard deviation.

Plankton samples from Dow Lake contained 11 species of rotifers, those from Lake Hope 3 species. *Keratella cochlearis*, the most common rotifer in Dow Lake, was followed in abundance by *Polyarthra vulgaris* Carlin, 1943. Other species were: *Asplanchna priodonta* Gosse, 1850; *Conochilus unicornis* Rousselet, 1892; *Kellicottia bostoniensis* (Rousselet, 1908); *K. longispina* (Kellicot, 1879); *Gastropus* sp.; *Lecane* sp.; *Monostyla* sp., *Synchaeta* sp.; and *Trichotria* sp. *Brachionus urceolaris*, the most common rotifer in Lake Hope, was followed in abundance by *Keratella cochlearis* and *Monostyla* sp. Identifications were based on Ahlstrom 1940, 1943; Edmondson 1959; Pennak 1953; and Rousselet 1902.

Rotifer species in samples from the 5 dates at Dow Lake ($\bar{X}=5.0\pm1.2$) were greater ($P < .01$) than those from Lake Hope ($\bar{X} = 2.0 \pm 1.2$), as determined by a means test. The numerical abundance of rotifers was also greater ($P < .02$) at Dow Lake ($\bar{X} = 221 \pm 158$) than at Lake Hope ($\bar{X} = 7.0 \pm 5.7$). Means for samples from specific date of collection are shown in figure 1, along with the proportion of Dow Lake rotifers represented by *Keratella cochlearis* and *Polyarthra vulgaris*.

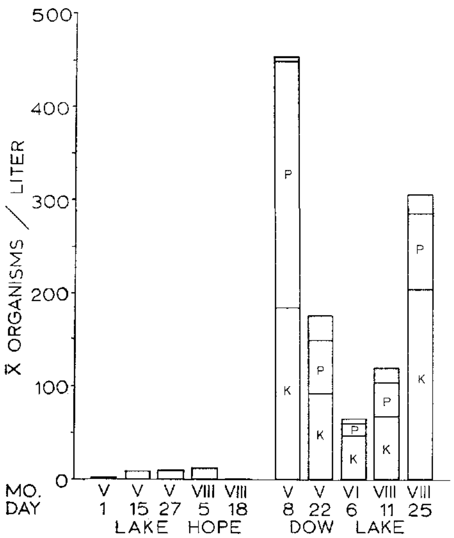

FIGURE 1. Summary of planktonic rotifer abundance, as mean number of organisms per liter, for each of 5 sampling dates in Lake Hope and Dow Lake. Proportions *Keratella cochlearis* (K) and *Polyarthra vulgaris* (P) are given for Dow Lake samples.

Species diversity (H') differed ($P < 0.05$) when based on pooled samples from the 5 dates: Dow Lake ($\bar{X} = 1.3 \pm 0.24$), Lake Hope ($\bar{X} = 0.46 \pm 0.64$). But the difference was even greater ($P < .001$), when based on individual samples: Dow Lake ($\bar{X} = 0.95 \pm 0.44$, n = 79), Lake Hope ($\bar{X} = 0.16 \pm 0.41$, n = 71).

Using the $S_{H'}$ similarity measure based on shared species diversity (Hummon 1974), rotifers from among pooled samples on 5 dates at Dow Lake shared 51.8% ± 3.9 of their diversity, whereas those at Lake Hope shared only 7.0% ± 3.4 of their diversity. More important, only 0.4% ± 0.1 was shared between permutations of the 2 sets of pooled samples. Using means tests, $S_{H'}$ values from among Dow Lake pools differed from those among Lake Hope pools ($P < .001$) and from those between Dow Lake and Lake Hope pools ($P < .001$), and $S_{H'}$ values from among Lake Hope pools differed from those between Dow Lake and Lake Hope pools ($P < .01$).

Tolerance tests indicated that the 24 hr lethal tolerance range (LT_{50}) for *Keratella cochlearis* from Dow Lake was pH 4.8 to 10.6, and for *Brachionus urceolaris* from Lake Hope it was pH 3.3 to 10.2. For *K. cochlearis*, 8% died over 24 hr in Dow Lake water and 50% in Lake Hope water, whereas 28% of *B. urceolaris* died in Lake Hope water but only 7% in Dow Lake water. Results of preference experiments were analysed for departure from randomness by a runs test. In every case there was a greater than random number of runs ($P < .001$).

DISCUSSION

Data from rotifer communities under mine acid conditions (Parsons 1968, and this study) indicate that the groups based on organic acidity, outlined by Myers (1931), are inappropriate. Further evidence is that while *Brachionus urceolaris* was considered by Myers as an alkaline water type, it has since been reported from organic- (Ahlstrom 1940) and mine- (Parsons 1968) acid water of pH 3.3 to 4.0. The pH levels of unpolluted natural waters are largely governed by the carbonate buffering system, hard carbonate-rich waters being alkaline and soft carbonate-poor waters being acidic. With the addition of mine acid, this equilibrium is disturbed and the buffering system destroyed, resulting in waters which may be both hard and acidic.

Based on late spring-late summer data, planktonic rotifers of Dow Lake exceeded those of Lake Hope in all categories analysed: species number, numerical abundance, species diversity, and faunal similarity. That these relationships are not characteristic of all faunal subcommunities was indicated by Orciari and Hummon (1975), working with benthic oligochaetes from these same 2 lakes. They found that only species diversity differed. While shared diversity among oligochaetes of Lake Hope did not differ from that among oligochaetes of Dow

Lake, both differed from that of permutations between samples from the 2 lakes, as found in our study.

Tolerance tests indicate that *K. cochlearis* and *B. urceolaris* were both eurytopic for hydrogen-ion concentration. Assuming otherwise-identical life cycles for these species and comparing the effect of Dow Lake and Lake Hope waters on their survivorships, selection pressures favor *B. urcheolaris* in both Dow Lake (1.1%) and Lake Hope (30.6%) waters. With respect to preference tests, neither species appeared sensitive to pH gradients, even when their numbers were being decimated by lethal alternative pH conditions. Rotifers of both species on encountering a pH gradient tended to make the same choice, to cross or not to cross, that they made on their last previous encounter, regardless of the water composition on either side of the gradient.

The picture that emerged was of more or less eurytopic species transported from one water body to another by physical or biological agents. On reaching a body such as Lake Hope, mine effluents appeared to act as a dominant physical control on abundance and diversity of the rotifer subcommunity, perhaps through changes in both birth and death rates. If this is the case, we hypothesize relatively high rates of species turnover in Lake Hope, correlated with high rates of colonization and extinction.

In Dow Lake, physical and chemical conditions appeared nearer optimal, with greater abundance and diversity of rotifers being regulated by cyclic physical-chemical factors or by biological accommodation, such as predation or competition. Ironically despite its tested compatibility to the physical-chemical conditions of Dow Lake, the dominant rotifer of Lake Hope, *Brachionus urceolaris*, was absent from all our Dow Lake samples. Sample error, biological interaction or some complex of factors probably best account for this observation.

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